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10/500,933	09/14/2005	Tong Shangguan	TRA-027.01	1561

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EXAMINER	
SCHNIZER, RICHARD A	
ART UNIT	PAPER NUMBER
1635	
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Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Office Action Summary	Application No.	Applicant(s)	
	10/500,933	SHANGGUAN ET AL.	
	Examiner	Art Unit	
	Richard Schnizer	1635	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE *3* MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *13 October 2009*.

2a)☒ This action is **FINAL**. 2b)☐ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *See Continuation Sheet* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1-7, 10, 11, 13-28, 30-34, 36-69, 71-80, 84, 85, 87-101, 104-107, 109-142, 144-151, and 153-168* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☐ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date *10/13/09*.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .
5) ☐ Notice of Informal Patent Application
6) ☐ Other: _____.

Continuation of Disposition of Claims: Claims pending in the application are 1-7, 10, 11, 13-28, 30-34, 36-69, 71-80, 84, 85, 87-101, 104-107, 109-142, 144-151, and 153-168 .

DETAILED ACTION

An amendment was received and entered on 10/13/09.

Claims 29, 30, 102, and 103 were canceled.

Claims 1-7, 10, 11, 13-28, 30-34, 36-69, 71-80, 84, 85, 87-101, 104-107, 109-142, 144-151, and 153-168 remain pending and are under consideration.

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

Claims 1-7, 10, 11, 13-17, 25-27, 31, 33, 34, 36-69, 71-80, 84-91, 99, 100, 104, 106, 107, 109-142, 144-147, and 153-168 stand rejected under 35 U.S.C. 103(a) as being unpatentable over Papahadjopoulos et al (US Patent 4,235,871), in view of Kikuchi et al (US 4,687,661), Meers et al (US Patent 6,120,797), Lenk et al (US Patent 5,169,637), and Thierry (US 6,096,335).

Papahadjopoulos taught a variety of liposomes comprising nucleic acids such as DNA or RNA. See column 3, lines 28-40; column 6, lines 31-43; column 8, lines 45-68 column 13, lines 59-67; and claims 16 and 17. The liposomes may consist of a fusogenic lipid such as dioleoylphosphatidylethanolamine (DOPE) or phosphatidylserine (PS), and may also comprise a variety of other lipids including cholesterol, see paragraph bridging columns 3 and 4. PS is a fusogenic lipid. The liposomes were from

200-400 nanometers in size (column 6, lines 11-14).

Papahadjopoulos taught a method of making liposomes by combining lipids as discussed above and nucleic acids in an inert solvent to form an emulsion, thereafter forming a gel, and finally converting the gel to a suspension of liposomes by addition of an aqueous medium. See entire document, especially e.g. claim 1, and column 4, line 45 to column 6, line 30.

Regarding claim 89, Papahadjopoulos taught addition of an aqueous solvent to the gel, rather than addition of the gel to an aqueous solvent, but this detail is considered to be a simple matter of design choice, and is therefore an obvious variant of the method of Papahadjopoulos.

Regarding claims 111-122 and 146, Papahadjopoulos is silent as to the total amount of lipid forming and fusogenic lipid expressed as a weight-percent of the gel, but the amounts of these lipids are considered to be result-effective variables that are obvious to optimize in order to modulate the characteristics of the resultant liposomes. See e.g. column 4, lines 53-58; and column 6, lines 11-18 and 28-34.

Regarding claims 124-135, Papahadjopoulos is silent as to the ratio of the weight of the increment of aqueous medium that can be used to wash the gel, and the weight of the gel itself. However, this is a parameter that would be routinely optimized by one of ordinary skill, see e.g. column 4, lines 53-58.

Regarding claims 136-140, Papahadjopoulos is silent as to the total amount of nucleic acid expressed as a weight-percent of the gel, but the amounts of the nucleic acid is clearly a result-effective variable that is routinely optimized by one of ordinary

skill. in order to modulate the characteristics of the resultant liposomes. See e.g. column 6, lines 11-18 and 28-34.

Instant claim 145 requires that the gel or gel particles lack a hydrating agent, and that no hydrating agent is used in step A of the method. The specification defines a hydrating agent at paragraph 74 as a compound having at least two ionizable groups, one of which ionizable groups is capable of forming an easily dissociative ionic salt, which salt can complex with the ionic functionality of the liposome-forming lipid. Papahadjopoulos exemplifies the use of buffers comprising histidine and TES, both of which appear to meet the definition of a "hydrating agent". However, Papahadjopoulos did not require that the aqueous phase added to the lipids must be buffered, but only that it comprises the biological agent to be encapsulated. The inclusion or exclusion of a buffering agent in the aqueous phase is a matter of design choice, as is the selection of any particular buffering agent. It would have been obvious to one of ordinary skill in the art to either include in, or exclude from, the aqueous phase an ionizable agent such as a buffer. If one chose to include a buffer, it would have been obvious to use any known biological buffer, including monoionic buffers such as imidazole.

Regarding claim 146, Papahadjopoulos is silent as to the total amount of organic solvent remaining in the gel, but it is clear that this is a result-effective variable that is routinely optimized by one of ordinary skill. See e.g. column 4, lines 53-58.

Papahadjopoulos did not teach a water miscible organic solvent or N-acyl phosphatidylethanolamines.

Kikuchi taught that the use of volatile organic solvents for dissolution of lipids in

methods of making liposomes was problematic because these organic solvents tend to remain in the final preparation and can be harmful to human health. Kikuchi suggests the use of water miscible organic solvents, such as ethylene glycol, for dissolution of lipids in methods of preparing liposomes. See column 1, line 60 to column 2, line 14; and column 2, lines 42-50. Kikuchi disclosed a number of acceptable organic solvents, including glycerol, ethylene glycol, and propylene glycol. Neither Papahadjopoulos nor Kikuchi specifically taught ethanol, methanol, or 2-propanol as water-miscible organic solvents.

Lenk taught a variety of solvents that could be used to solubilize lipids, including ethanol, methanol, or 2-propanol. See table VI at column 40.

Thierry taught methods of making lipid-nucleic acid complexes, wherein lipids are solubilized in an alcohol having 1-4 carbon atoms, preferably 2 to 3 (such as ethanol, isopropanol, glycerol, ethylene glycol, or propylene glycol), a nucleic acid is added to the lipids, and complexes are formed comprising the nucleic acids encapsulated in a lamellar spherical vesicle. See Fig. 3; and column 8, lines 20-67). DNAs, RNAs, plasmids, antisense oligonucleotides, and ribozymes are examples of nucleic acids that can be encapsulated. See paragraph bridging columns 6 and 7; column 9, lines 16-22; paragraph bridging columns 14 and 15; column 15, lines 51-58; and claim 29. Thierry also suggested the use of DOPE in the complexes. See e.g. column 5, lines 51-64.

Meers taught N-acyl phosphatidylethanolamines, including a N-dodecanoyl dioleoyl phosphatidylethanolamine, for use in liposome formation. See e.g. abstract; and column 4, lines 40-51. Meers also suggested the use of DOPE and DOPC in

liposomes. See column 5, line 51 to column 6, line 1.

It would have been obvious to one of ordinary skill in the art at the time of the invention to substitute the water miscible solvent of Kikuchi in the method of Papahadjopoulos because Kikuchi taught that the use of water miscible organic solvents was safer for applications in which the product liposomes were to be used in vivo. It would have been similarly obvious to use the lipids of Meers in the liposomes of Papahadjopoulos because Meers suggested their use for liposome formation, and because N-dodecanoyl dioleoyl phosphatidylethanolamine and DOPE are fusogenic. See e.g. column 1, lines 48-60. Finally it would have been obvious to encapsulate plasmids or antisense oligonucleotides because this is suggested by Thierry.

It would have been obvious to one of ordinary skill in the art at the time of the invention to use in the invention of Papahadjopoulos any water miscible organic solvent that can be used to solubilize liposome-forming lipids in view of the teachings of Kikuchi, as discussed above. It would have been obvious to use ethanol or 2-propanol because both Lenk and Thierry taught that these can be used to solubilize lipids. Note also that MPEP 2144.06 indicates that when it is recognized in the art that elements of an invention can be substituted, one for the other, while retaining essential function, such elements are art-recognized equivalents. An express suggestion to substitute one equivalent component or process for another is not necessary to render such substitution obvious. Furthermore, MPEP 2144.07 indicates that the selection of a known material based on its suitability for its intended use supports the determination of prima facie obviousness. In this case it was clear that the glycerol, ethylene glycol, and

propylene glycol of Kikuchi were art recognized equivalents of the ethanol and 2-propanol of Meers because each was disclosed as one of several alternative lipid solvents by Thierry.

Thus the invention as a whole was prima facie obvious.

Claims 18-24, 28, 32, 92-98, 101, 105, and 148-151 stand rejected under 35 U.S.C. 103(a) as being unpatentable over Papahadjopoulos, Kikuchi, Meers, Lenk, and Thierry (US 6,096,335), as applied to claims 1-7, 10, 11, 13-17, 25-27, 31, 33, 34, 36-69, 71-80, 84-91, 99, 100, 104, 106, 107, 109-142, 144-147, and 153-168 above, and further in view of Eppstein et al (US Patent 4897355) taken with the evidence of GenBank Accession No. M77788.

The teachings of Papahadjopoulos, Kikuchi, Meers, Lenk, and Thierry are summarized above and render obvious methods of making liposomes comprising nucleic acids and N-acyl phosphatidylethanolamine lipids by solubilizing liposome-forming lipids in a C_1-C_3 alcohol.

These references are silent as to the sizes of the plasmid DNAs and oligonucleotides, and as to transfection incubation temperature and in vivo administration of plasmids.

Eppstein taught that liposomes could be used to encapsulate and deliver to cells plasmid DNAs and oligonucleotides, including pSVCAT (5 kbp) and oligonucleotides of an average length of about 130 bp. See column 3, lines 56-59; column 8, lines 32-40 and 43-45; column 10, lines 56-59; column 48 lines 24-50, and paragraph bridging

columns 48 and 49. GenBank Accession No. M77788 provides evidence that PSVCAT is 5003 base pairs in length.

Eppstein also taught a variety of lipids that could be used to form liposomes including dioleoylphosphatidylcholine (DOPC), dipalmitoylphosphatidylcholine (DPPC), distearoylphosphatidylcholine, and cholesterol (see paragraph bridging columns 7 and 8; column 16, lines 53-and 54; and column 38, line 37).

Eppstein also taught methods of transfecting eukaryotic cells in vitro at 37^{o}C (column 45, lines 43-52), as well as intravenous delivery to humans (see column 8, lines 1-13; column 10, lines 37-62; column 12, lines 48-53; column 13, lines 27-29; and column 20, lines 21-41).

It would have been obvious to one of ordinary skill in the art at the time of the invention to use the method of Papahadjopoulos as modified by Kikuchi, Meers, Lenk, and Thierry to encapsulate the plasmid or oligonucleotides of Eppstein because Papahadjopoulos suggests that the liposomes will protect nucleic acids from degradation (see e.g. column 13, lines 59-67), and because one of ordinary skill would clearly appreciate their utility for this purpose in view of the teachings of Eppstein. It would have been similarly obvious to use the lipids of Eppstein in the methods of Papahadjopoulos, because MPEP 2144.07 indicates that the selection of a known material based on its suitability for its intended use supports the determination of prima facie obviousness. In this case the lipids of Eppstein are clearly suitable for making liposomes. Finally it would have been obvious to use the liposomes to transfect

eukaryotic cells in vitro or in vivo, as well as for intravenous delivery in humans because this was suggested by Eppstein.

Response to Arguments

Applicant's arguments filed 10/13/09 have been fully considered but they are not persuasive.

Applicant argues that argues at Papahadjopoulos “differs from the present liposomes and methods in that an entirely different class of organic solvent is used, i.e., solvents with no aqueous solubility versus C1-C3 alcohols, the gel contains primarily an aqueous phase and lipids, instead of an alcohol, and the lipids are not fusogenic." The Office agrees but notes that Papahadjopoulos was not relied upon to teach these limitations, instead they are rendered obvious by combination with the Kikuchi, Lenk, Meers, and Thierry references. Applicant asserts that such a combination could only arrive at such a combination by hindsight. However, it must be recognized that any judgment on obviousness is in a sense necessarily a reconstruction based upon hindsight reasoning. But so long as it takes into account only knowledge which was within the level of ordinary skill at the time the claimed invention was made, and does not include knowledge gleaned only from the applicant's disclosure, such a reconstruction is proper. See MPEP 2145 (IX)(A). Applicant has not pointed to any claim limitation that was met by using knowledge gleaned only from the applicant's disclosure, and not the prior art.

Applicant asserts that the present claims differ in that the liposomes are directly formed by adding an aqueous medium to a gel where the gel contains a C1-C3 alcohol, the lipids, and the compound to be encapsulated. However, one of ordinary skill in the art aware of the teachings of Kikuchi would have been motivated to substitute a water miscible solvent for the organic solvent of Papahadjopoulos in order to avoid having potentially harmful organic solvents remaining in the final preparation because Kikuchi explicitly suggested the substitution of water miscible solvents for organic solvents for this very reason. Further, one of ordinary skill aware of the teachings of Lenk would have understood that water miscible solvents appropriate for dissolving lipids included ethanol, methanol, or 2-propanol. Combination of the lipid solution in C1-C3 alcohol with an aqueous solution comprising a nucleic acid would result in gel formation, and subsequent addition of an aqueous medium would convert the gel to a suspension of liposomes as taught by Papahadjopoulos.

Applicant argues that there is no rational basis for a person of skill in the art to modify the references with any reasonable expectation of success. Applicant notes that Paphadjopoulos requires the use of water immiscible solvents for forming inverted micelles and emphasizes that removal of these solvents prior to the final dissolution in an aqueous phase is "essential for high capture percentage in this method and is a critical difference between the process of our invention and all previous methods described." The Examiner agrees that Papahadjopoulos indicated the importance of forming inverted micelles in an emulsion of a water immiscible solvent and an aqueous phase. However, it is also clear that those of ordinary skill in the art understood that this

was not necessary for encapsulation of nucleic acids into liposomes, and that less toxic organic solvents could be used to dissolve lipids, such as those suggested by Kikuchi or Lenk. Note that Kikuchi taught methods of making liposomes by dissolving lipids in organic solvents to produce a liquid, paste, or gel, adding an aqueous phase comprising a nucleic acid, and stirring to form liposomes comprising the nucleic acid. Kikuchi found that homogeneous liposomes can be obtained with ease and good reproducibility when membrane components forming liposomes are dissolved or swollen in a water-soluble and physiologically acceptable non-volatile organic solvent and the resulting mixture is further mixed and stirred with an aqueous medium. See column 1, line 62 to column 2, line 2. There is no evidence of record that use of a water miscible solvent in the method of Papahadjopoulos would render the method inoperative. Based on the teachings of Papahadjopoulos cited by Applicant, one might expect to see a decrease in encapsulation yield, but this is not evidence of inoperability. Further, in performing the substitution, one would gain the advantage of eliminating from the final product organic solvents that are harmful to human health, as taught by Kikuchi. Accordingly, there is a rational basis for substituting the water miscible solvent for the water immiscible solvent. In view of these teachings, and the teachings of Lenk and Thierry that lipids could be dissolved in C_1-C_3 alcohols, one of ordinary skill would clearly have had a reasonable expectation of success in using C_1-C_3 alcohols as an organic solvent in the methods of either Papahadjopoulos of Kikuchi. One would have realized that formation of an emulsion is not required for encapsulation, and that the use of toxic organic solvents could be avoided.

Applicant also argues that one of ordinary skill would not have used C_1-C_3 alcohols as an organic solvent because one appreciates that nucleic acids are not soluble in these solvents, relying for support on passages from "Molecular Cloning: A Laboratory Manual. However, this reference clearly teaches that the precipitation of nucleic acids in alcohols is performed with the addition of salt solutions. These salts decrease the solubility of the nucleic acid in water, and facilitate precipitation. The cited art does not teach the addition of salts that would decrease the solubility of nucleic acids such that they would precipitate in C_1-C_3 alcohols. The art cited by Applicant also teaches the use of at 2-3 volumes of alcohol for precipitation from a given volume of aqueous nucleic acid solution, whereas Kikuchi indicates that water miscible organic solvents can be used at about 0.001 to about 2 parts by weight per part by weight of the aqueous medium. Accordingly, one would have had reason to use a volume of alcohol that was less than what is generally used to precipitate nucleic acids, e.g. about 0.001 to about 2 parts by weight per part by weight of the aqueous medium. Finally, Thierry provides objective evidence that one of ordinary skill in the art prior to the invention could have used the claimed alcohols in combination with nucleic acids to form liposomes encapsulating the nucleic acids. See e.g. Example 6 at column 13.

Applicant asserts that the reference plainly states that states that nucleic acids are insoluble in ethanol, and indicates the use of ethanol to wash precipitated nucleic acids. The Examiner agrees that nucleic acids are insoluble in concentrated solutions of ethanol. That's why ethanol is used to precipitate nucleic acids. As discussed above, a salt is generally used to trigger precipitation. Once the nucleic acid is

precipitated, it can be washed with 70-95% ethanol without fear of it redissolving rapidly. This has absolutely no bearing on the issue of obviousness of the claimed method. Kikuchi taught that water miscible organic solvents can be used at about 0.001 to about 2 parts by weight per part by weight of the aqueous medium. This is below the range used to precipitate, or wash, nucleic acids, which is 2-3 volumes of ethanol. In any case, as evidenced in *Molecular Cloning*, one of ordinary skill at the time of the invention was well aware of the solubility characteristics of nucleic acids, and would have known better than to use an amount of ethanol or propanol that would result in precipitation instead of liposome formation.

Applicant points out that Thierry taught methods of making compositions that do not appear to be classical liposomes in the sense that they are not lipid bilayer vesicles. Applicant concludes that Thierry fails to describe formation of liposomes using alcohols. This is unpersuasive regarding a lack of obviousness, because Thierry is relied upon to teach that lipids of the type frequently used to form liposomes are soluble in C1-C3 alcohols. It is immaterial whether or not the actual structures formed by Thierry are lipid bilayer vesicles or not. Note also that Thierry provides objective evidence that one can solubilize lipids in C1-C3 alcohols and combine them with nucleic acid solutions without precipitation of the nucleic acids.

Finally Applicant argues that one of skill would not have been motivated to use C1-C3 alcohols to encapsulate [nucleic acids] as claimed, because Lenk used these in the process of encapsulating small organic molecules. This is unpersuasive because Lenk merely provides the information that C1-C3 alcohols are useful for solubilizing

lipids. Motivation to use water miscible solvents including some alcohols, instead of organic solvents, comes from Kikuchi.

For these reasons the rejection is maintained.

Conclusion

No claim is allowed.

THIS ACTION IS MADE FINAL. Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the date of this final action.

Any inquiry concerning this communication or earlier communications from the examiner(s) should be directed to Richard Schnizer, whose telephone number is 571-272-0762. The examiner can normally be reached Monday through Friday between the hours of 6:00 AM and 3:30 PM. The examiner is off on alternate Fridays, but is sometimes in the office anyway.

If attempts to reach the examiner by telephone are unsuccessful, the Examiner's supervisor, Tracy Vivlemore, can be reached at (571) 272-0763. The official central fax number is 571-273-8300. Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to (571) 272-0547.

Patent applicants with problems or questions regarding electronic images that can be viewed in the Patent Application Information Retrieval system (PAIR) can now contact the USPTO's Patent Electronic Business Center (Patent EBC) for assistance. Representatives are available to answer your questions daily from 6 am to midnight (EST). The toll free number is (866) 217-9197. When calling please have your application serial or patent number, the type of document you are having an image problem with, the number of pages and the specific nature of the problem. The Patent Electronic Business Center will notify applicants of the resolution of the problem within 5-7 business days. Applicants can also check PAIR to confirm that the problem has been corrected. The USPTO's Patent Electronic Business Center is a complete service center supporting all patent business on the Internet. The USPTO's PAIR system provides Internet-based access to patent application status and history information. It also enables applicants to view the scanned images of their own application file folder(s) as well as general patent information available to the public.

For all other customer support, please call the USPTO Call Center (UCC) at 800-786-9199.

/Richard Schnizer/
Primary Examiner, Art Unit 1635